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Presynaptic Proteoglycans: Sweet Organizers of Synapse Development

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Synaptic adhesion molecules control neuronal synapse development. In this issue of *Neuron*, Siddiqui et al. (2013) and de Wit et al. (2013) demonstrate that LRRTM4, a postsynaptic adhesion molecule, *trans*-synaptically interacts with presynaptic heparan sulfate proteoglycans (HSPGs) to promote synapse development.

One of the key mechanisms underlying the development of neuronal synapses and connectivity in the nervous system is synaptic adhesion. Synaptic adhesion molecules are thought to regulate diverse steps of synaptic development, including the formation, maturation, maintenance, and plasticity of neuronal synapses. A well-known example of such synaptic adhesion proteins are the neuroligins, which *trans*-synaptically interact with presynaptic neurexins (Südhof, 2008). A series of studies on this interaction has significantly contributed to our current understanding of the mechanisms underlying synapse development, synaptic transmission, and plasticity and of neuropsychiatric diseases such as autism spectrum disorders.

However, given the enormous diversity of neuronal connectivity in the nervous system, it is not surprising that a large number of synaptic adhesion molecules exist. Important examples of such molecules include neurexin-binding postsynaptic adhesion molecules such as leucine-rich repeat transmembrane neuronal proteins (LRRTMs) and Cbln-GluRδ (Williams et al., 2010) and LAR family receptor protein tyrosine phosphatase (LAR-PTP)-binding postsynaptic adhesion molecules such as NGL-3, TrkC, IL1RAPL1, and Slitrks (Takahashi and Craig, 2013) (Figure 1).

The LRRTMs are a family of postsynaptic adhesion molecules with four known members. All contain leucine-rich repeats in the extracellular region, a single transmembrane domain, and a cytoplasmic region containing a C-terminal PDZ-binding motif that is required for binding to the postsynaptic scaffolding protein PSD-95 (Laurén et al., 2003) (Figure 1). All four LRRTMs are capable of inducing presynaptic differentiation in contacting axons (Linhoff et al., 2009), indicating that they interact with specific presynaptic ligands. Indeed, LRRTM1 and LRRTM2 *trans*-synaptically interact with neurexins, and these interactions promote excitatory synapse development in a bidirectional manner (de Wit et al., 2009; Ko et al., 2009; Siddiqui et al., 2010). However, it has remained unclear whether different LRRTMs interact with distinct binding partners to promote presynaptic development.

Two papers on LRRTM4 reported in this issue of *Neuron* (de Wit et al., 2013; Siddiqui et al., 2013) show that this is the case. These studies demonstrate that LRRTM4 proteins are particularly abundant in the molecular layers of the hippocampal dentate gyrus (DG) and that postsynaptic LRRTM4 *trans*-synaptically interacts with presynaptic membrane-associated heparan sulfate proteoglycans (HSPGs), such as glypicans and syndecans (Figure 1). These interactions are HS dependent and promote excitatory, but not inhibitory, synapse development in a bidirectional manner. Knockdown of LRRTM4 in cortical pyramidal neurons by in utero electroporation reduces dendritic spine number and synaptic levels of AMPA-type glutamate receptors (AMPARs) (de Wit et al., 2013). Moreover, mutant mice that lack LRRTM4 show reductions in the density of dendritic spines and frequency of miniature excitatory postsynaptic currents in the DG but not CA1 region of the hippocampus (Siddiqui et al., 2013). Intriguingly, LRRTM4-deficient neurons show impaired activity-dependent trafficking of AMPARs (Siddiqui et al., 2013), indicating that LRRTM4 may regulate synaptic plasticity.

These results show that different LRRTMs display distinct cell-type- and pathway-specific expression patterns and induce presynaptic differentiation through their specific ligands. The new studies also demonstrate that HSPG clustering on axonal surfaces promotes presynaptic differentiation, a function distinct from that of glypicans 4 and 6, which are secreted from astrocytes and promote synaptic AMPAR clustering and excitatory synapse development in retinal ganglion cells (Allen et al., 2012). The new results further indicate that LRRTM4 regulates basal and activity-dependent synaptic localization of AMPARs, consistent with the reported biochemical association of LRRTM4 with AMPARs (Schwenk et al., 2012).

Although these findings provide significant insights into the molecular and cellular mechanisms underlying the development of neuronal connectivity, a host of unanswered questions remain. First, it is unclear exactly how negatively charged HS moieties are required for LRRTM4-dependent presynaptic differentiation; they may regulate the strength of adhesions or cell-surface turnover of ligands. If HS is an important determinant of presynaptic development, would secreted forms of HSPGs from neighboring cells compete with presynaptic

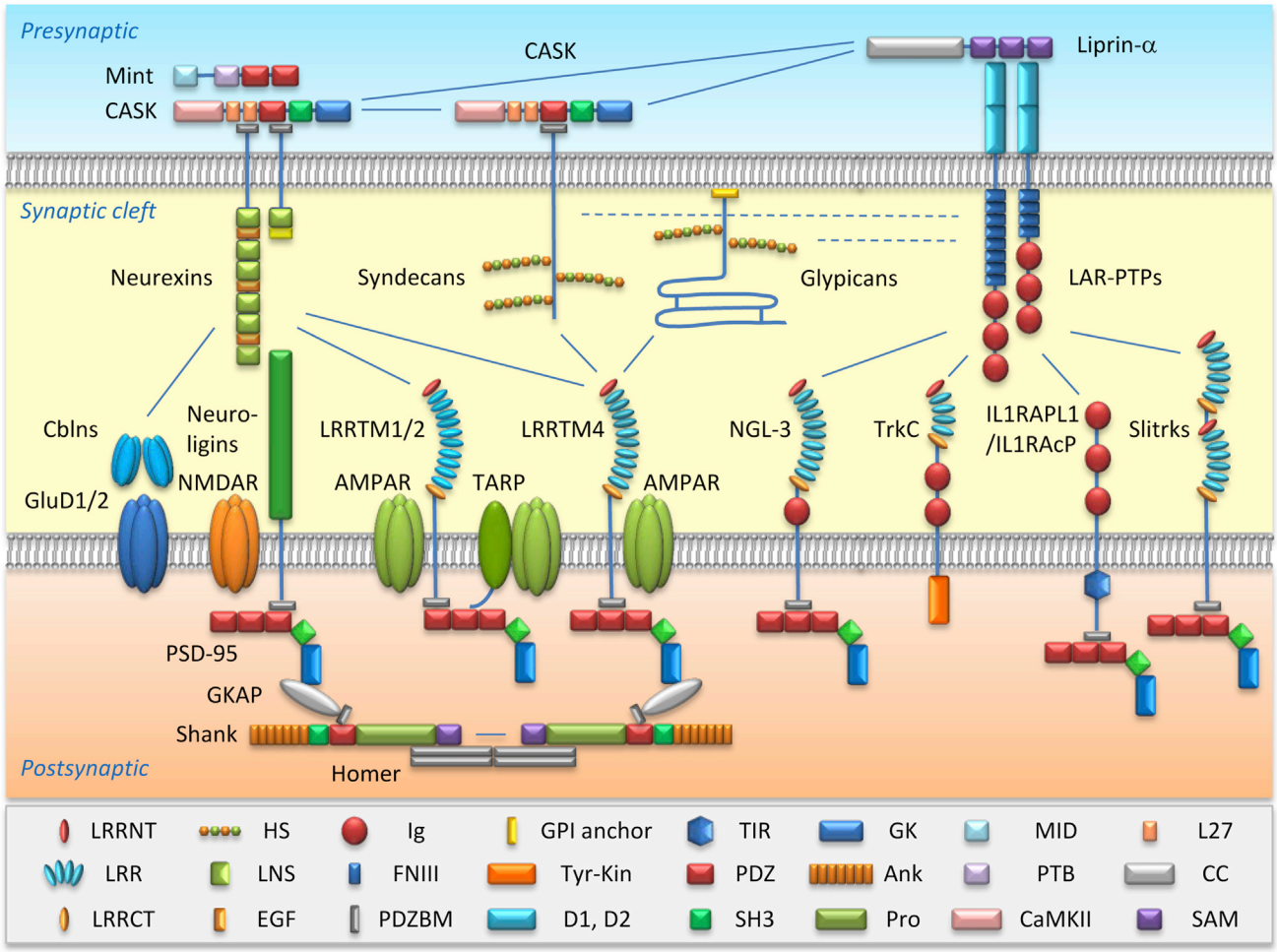

Figure 1. Examples of *trans*-Synaptic Interactions Known to Regulate Excitatory Synapse Development
The diagram depicts two relatively well-known groups of *trans*-synaptic adhesion complexes involving presynaptic neurexins and LAR-PTPs and an emerging group involving presynaptic HSPGs including glypicans and syndecans. Known protein-protein interactions are indicated by the direct contacts of proteins or solid lines. Dotted lines indicate potential interactions, based on *Drosophila* protein-protein interactions.

HSPGs and modulate LRRTM4-induced presynaptic differentiation? In addition, HSPGs, including glypicans and syndecans, show widespread expression patterns in the brain, in contrast to the preferential expression of LRRTM4 in the DG. Therefore, non-DG brain regions may have other types of postsynaptic ligands for HSPGs.

Glypicans are glycosyl-phosphatidyl inositol (GPI)-anchored HSPGs that lack cytoplasmic regions, unlike syndecans. Given that neurexins and LAR-PTPs interact with cytoplasmic proteins to promote presynaptic development (Südhof, 2008; Takahashi and Craig, 2013), glypicans may interact in a *cis* manner with as yet unknown coreceptors containing transmembrane and cytoplasmic domains. Prime candidates for such coreceptors are LAR-PTPs because Dally-like, a *Drosophila* glypican, interacts with dLAR (Johnson et al., 2006). Given that LAR-PTPs possess a membrane-proximal tyrosine phosphatase (D1) domain in addition to the membrane-distal and catalytically inactive protein-protein interaction (D2) domain, glypicans may also form a signal-transducing complex with LAR-PTPs. In addition, because LAR and neurexins probably act together through shared cytoplasmic proteins to promote presynaptic development (Takahashi and Craig, 2013), HSPGs may functionally cooperate with both LAR-PTPs and neurexins (Figure 1). This cooperation may also involve the *trans*-synaptic interaction of LRRTM4 with neurexins (de Wit et al., 2013), although this interaction was not detected in the other study (Siddiqui et al., 2013).

LRRTM4 regulates basal and activity-dependent synaptic localization of AMPARs, similar to the reported LRRTM1/2-dependent regulation of AMPAR-mediated excitatory synaptic transmission (de Wit et al., 2009; Ko et al., 2011; Soler-Llavina et al., 2011) and synaptic stabilization of newly inserted AMPARs during long-term potentiation (LTP) (Soler-Llavina et al., 2013). The details of how LRRTM4 mediates these regulatory functions remain unclear. Does LRRTM4 directly interact with and promote surface expression and synaptic localization of AMPARs, similar to LRRTM1/2 (de Wit et al., 2009; Soler-Llavina et al., 2011)

and also transmembrane AMPA receptor regulatory proteins (TARPs) (Jackson and Nicoll, 2011)? Does LRRTM4 affect the gating and pharmacological properties of AMPARs and modulate synaptic plasticity (i.e., LTP and long-term depression)? Does the *trans*-synaptic interaction between LRRTM4 and HSPGs, an adhesive feature absent in TARPs, have any role in AMPAR regulation, similar to the requirement of neurexin binding for LRRTM1/2-dependent synaptic stabilization of AMPARs during LTP (Soler-Llavina et al., 2013)? What sort of interplay exists between LRRTM4, LRRTM1/2, and TARPs—all of which are expressed in the DG—in AMPAR regulation? Do they compete or cooperate with each other?

Lastly, copy number variations of LRRTM4 and glypican 6 have been associated with autism spectrum disorders (Pinto et al., 2010). In addition, mutant mice that lack *Ext1*, which encodes an enzyme essential for HS biosynthesis, display autistic-like behaviors including impaired social interaction, reduced ultrasonic vocalizations, and repetitive behaviors (Irie et al., 2012). Whether and how deficits in LRRTM4-HSPG interactions contribute to the development of these disorders remain open questions.

In conclusion, the two new studies demonstrate that postsynaptic LRRTM4 *trans*-synaptically interacts with presynaptic HSPGs to promote excitatory synapse development, identifying HSPGs as an unexpected group of presynaptic organizers. Fruitful avenues for future research include determining whether similar HSPG-dependent *trans*-synaptic adhesions are widespread in various brain regions. If so, this would reinforce the importance of this newly emerging group of presynaptic organizers.

REFERENCES

Allen, N.J., Bennett, M.L., Foo, L.C., Wang, G.X., Chakraborty, C., Smith, S.J., and Barres, B.A. (2012). Nature *486*, 410–414.

de Wit, J., Sylwestrak, E., O'Sullivan, M.L., Otto, S., Tiglio, K., Savas, J.N., Yates, J.R., 3rd, Comoletti, D., Taylor, P., and Ghosh, A. (2009). Neuron *64*, 799–806.

de Wit, J., O'Sullivan, M.L., Savas, J.N., Condomitti, G., Caccese, M.C., Vennekens, K.M., Yates, J.R., III, and Ghosh, A. (2013). Neuron *79*, this issue, 696–711.

Irie, F., Badie-Mahdavi, H., and Yamaguchi, Y. (2012). Proc. Natl. Acad. Sci. USA *109*, 5052–5056.

Jackson, A.C., and Nicoll, R.A. (2011). Neuron *70*, 178–199.

Johnson, K.G., Tenney, A.P., Ghose, A., Duckworth, A.M., Higashi, M.E., Parfitt, K., Marcu, O., Heslip, T.R., Marsh, J.L., Schwarz, T.L., et al. (2006). Neuron *49*, 517–531.

Ko, J., Fuccillo, M.V., Malenka, R.C., and Südhof, T.C. (2009). Neuron *64*, 791–798.

Ko, J., Soler-Llavina, G.J., Fuccillo, M.V., Malenka, R.C., and Südhof, T.C. (2011). J. Cell Biol. *194*, 323–334.

Laurén, J., Airaksinen, M.S., Saarma, M., and Timmusk, T. (2003). Genomics *81*, 411–421.

Linhoff, M.W., Laurén, J., Cassidy, R.M., Dobie, F.A., Takahashi, H., Nygaard, H.B., Airaksinen, M.S., Strittmatter, S.M., and Craig, A.M. (2009). Neuron *61*, 734–749.

Pinto, D., Pagnamenta, A.T., Klei, L., Anney, R., Merico, D., Regan, R., Conroy, J., Magalhaes, T.R., Correia, C., Abrahams, B.S., et al. (2010). Nature *466*, 368–372.

Schwenk, J., Harmel, N., Brechet, A., Zolles, G., Berkefeld, H., Müller, C.S., Bildl, W., Baehrens, D., Hüber, B., Kulik, A., et al. (2012). Neuron *74*, 621–633.

Siddiqui, T.J., Pancaroglu, R., Kang, Y., Rooyakkers, A., and Craig, A.M. (2010). J. Neurosci. *30*, 7495–7506.

Siddiqui, T.J., Tari, P.K., Connor, S.A., Zhang, P., Dobie, F.A., She, K., Kawabe, H., Wang, Y.T., Brose, N., and Craig, A.M. (2013). Neuron *79*, this issue, 680–695.

Soler-Llavina, G.J., Fuccillo, M.V., Ko, J., Südhof, T.C., and Malenka, R.C. (2011). Proc. Natl. Acad. Sci. USA *108*, 16502–16509.

Soler-Llavina, G.J., Arstikaitis, P., Morishita, W., Ahmad, M., Südhof, T.C., and Malenka, R.C. (2013). Neuron *79*, 439–446.

Südhof, T.C. (2008). Nature *455*, 903–911.

Takahashi, H., and Craig, A.M. (2013). Trends Neurosci. Published online July 9, 2013. http://dx.doi.org/10.1016/j.tins.2013.06.002.

Williams, M.E., de Wit, J., and Ghosh, A. (2010). Neuron *68*, 9–18.